

Mechanical design of pollen apertures

(on a paper by Božič and Šiber, PNAS 2020)

Beginning of april is perhaps the most spectacular part of the year, when cherries bloom and the tree crowns become white from all the flowers. Japanese people expect with a special excitement this period of bloom - *sakura* - and people meet below cherry trees and enjoy in food and drink.

Although we see it in perhaps the most beautiful form when cherries bloom, the bloom and the flowers are everywhere, though people typically do not know much about the flowers of pumpkin, eggplant or potato. Flowering plants are not only the most conspicuous part of the plant kingdom, they are also the main source of food for humans and animals. Cereals, potatoes, tomatoes, legumes, pumpkins, cabbage, apples, almonds are only some of the flowering plants on whose fertility we fundamentally depend.



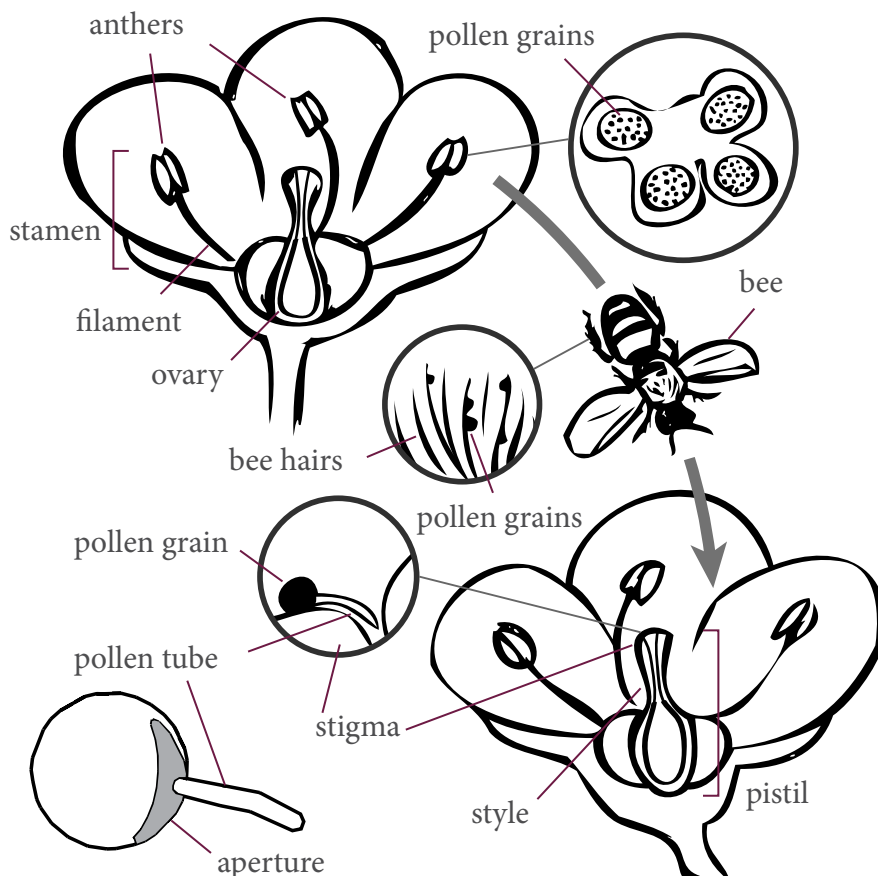
Prunus serrulata

We usually eat fruits which develop from flowers, but in some plant species we actually eat flowers. Such is the case e.g. with broccoli, cauliflower, artichoke, and capers, but many flowers which are rarely eaten are edible, e.g. flowers of mint, hibiscus, rose, violet...

The flower is a reproductive system of a plant and typically contains the organs of both sexes i.e. **pistil** (female part) and **stamens** (male part). In the fertilisation/pollination process, the pollen grains which are in the **anthers** get transported to the **stigma** and that is how the male genetic information from one plant mixes with the female information from another plant.

Transfer of a pollen grain from an **anther** of one plant to a stamen of another plant can be mediated by wind or water, but about 80% of flowering plants rely on pollinators, i.e. animals which perform the process. Most famous

of these animals are bees, but there are also bumblebees, flies, butterflies, wasps, ants, and pollinators need not be only insects – some plants are pollinated by colibri, sunbirds and bats, and in small number of cases even geckos and rodents. When a pollen grain lands on a stigma it starts to “germinate”, i.e. a pollen tube starts to grow on a particularly soft region of the grain wall called the **aperture**. The tube grows until it passes through the **style** and reaches the **ovary**. A gene transfer from the pollen grain to the ovule then takes place.

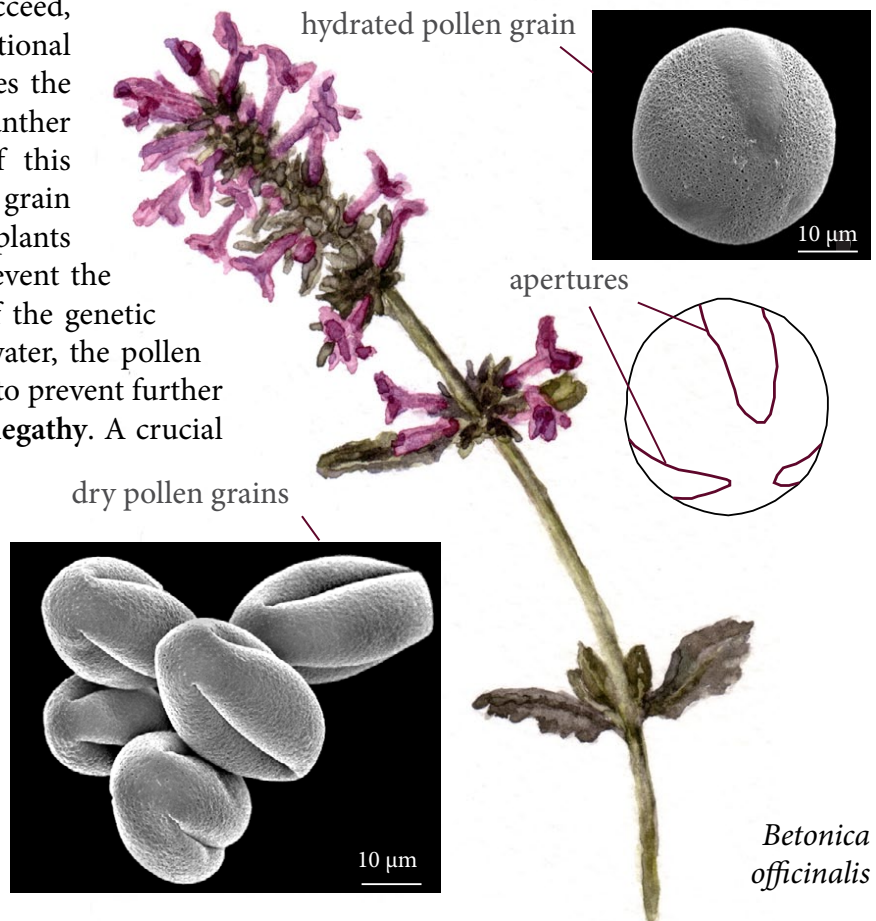


Huge almond orchards in California are pollinated by bees transported by beekeepers from all over the USA. Without these bees the almond crop would be much smaller. A large number of bees die during the pollination from the insecticides used to treat the almonds*.

* A.Wade et al, Insects 10, 20 (2019)

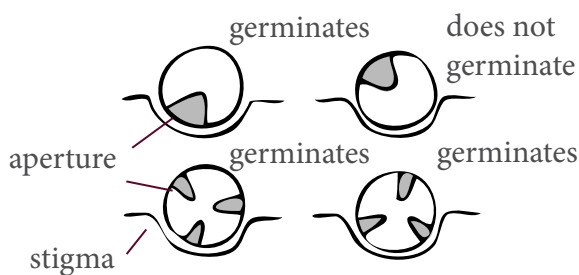
In order for the fertilization to succeed, the pollen grain must remain functional during the transition, i.e. when it leaves the environment of the flower and the anther from which it originates. Outside of this environment the pollen dries, and as the grain protects sensitive genetic material, the plants have developed mechanisms which prevent the drying of the grain and destruction of the genetic information it carries. When it loses water, the pollen grain typically “infolds” i.e. deforms so to prevent further drying – this process is called **harmomegathy**. A crucial role in the process is played by the parts of the grain wall where the pollen tube growth initiates –the apertures. The apertures enable infolding of the grain and prevent its crushing when it dries, but also enable its unfolding once it gets in the humid environment, i.e. on the stigma.

A scientific discipline investigating pollen of the existing but also extinct pollen species is called **palynology**.



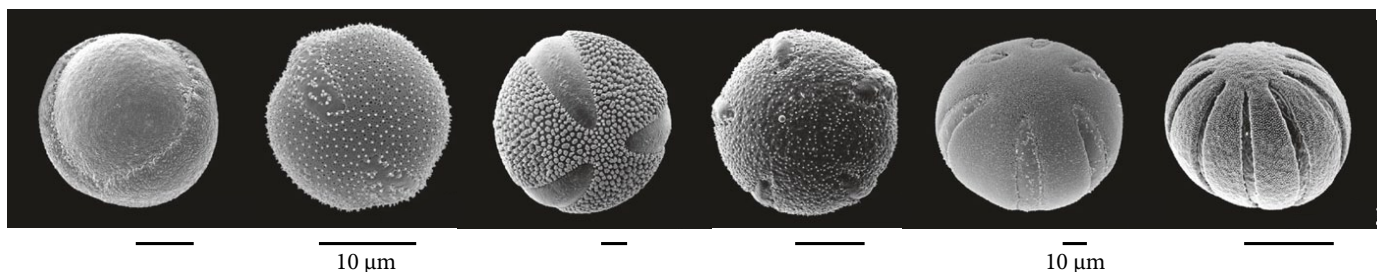
Betonica officinalis

The two largest groups of flowering plants – **monocots** and **eudicots** – differ in the number of apertures. Evolutionary older monocots (i.e. orchid, wheat and palm) have typically grains with one aperture, while younger and more numerous eudicots (e.g. sunflower, apple, maple) have three. Even older plants, **gymnosperms** (e.g. cypress, yew, ginkgo), have pollen grains with a single aperture or without clearly visible apertures. It is thus likely that the increase of the number of apertures is an evolutionary trend and that there is a reason for it.



When a grain lands on the stigma, it is necessary for the aperture to face the stigma in order for the germination of the pollen tube to initiate. With the same total aperture area, the probability of germination increases with the number of apertures. The question is, however, whether the increased number of the apertures influences the infolding of the pollen grain and its protection from the desiccation.

Pollen grains with 1, 2, 3, 5, 6 and 9 apertures



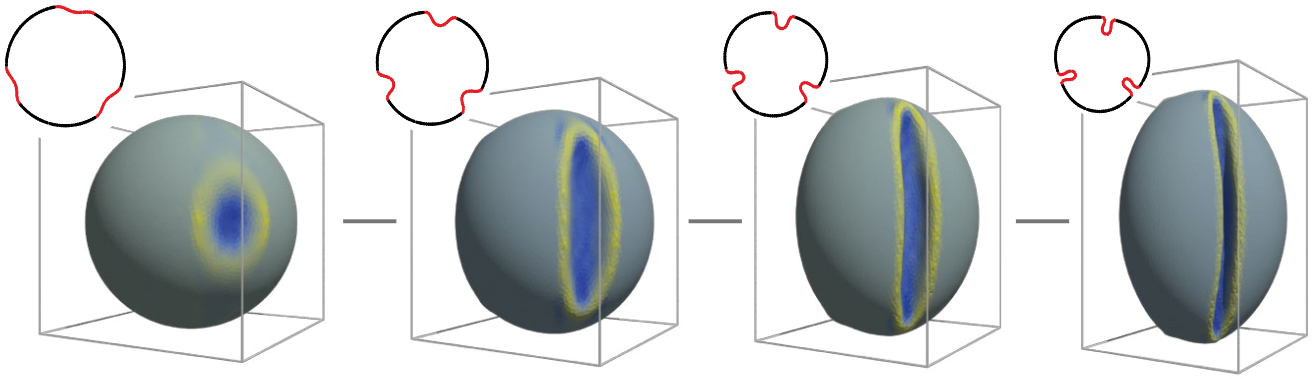
Božič and Šiber* investigated how the infolding of the pollen grain depends on its mechanical design i.e. on the size, shape and the number of apertures and on the elasticity of the grain wall. Although it was already known** that the apertures must be softer parts of the grain wall in order to enable and initiate the infolding, Božič and Šiber have shown in the elastic model of the grain that the apertures must be 15 to 100 times softer than the hard part of the wall in order for the infolding to proceed regularly and that it ends in the closure of the apertures, preventing thus further desiccation.

Size of the pollen grains depend on the species in question. Grain diameters can be smaller than 10 µm (e.g. in forget-me-not), but also larger than 150 µm (e.g. in pumpkin). The 10 µm length is denoted on the microscopic images of the grains (above) for each grain.

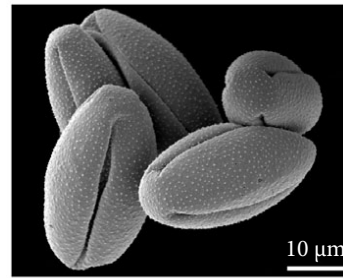
* A. Božič and A. Šiber, PNAS (2020), <https://doi.org/10.1073/pnas.2011084117>

** E. Katifori et al, PNAS 107, 7635 (2010)

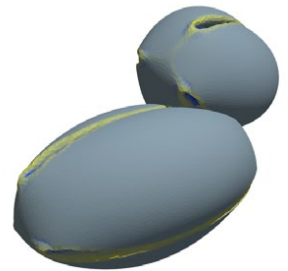
Theoretical calculation of the process of a regular closure of the pollen grain. The grain elongates as the apertures close.



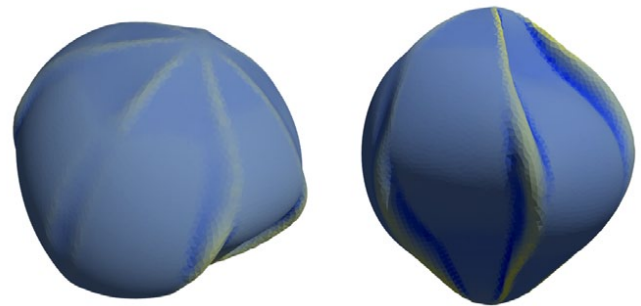
They also discovered that the infolding does not necessarily lead to the closure of the apertures – a deformation of the hard part of the pollen wall may occur which leaves the apertures open and does not prevent a further collapse of the grain.



Microscope images of pollen (left) and calculated shapes (right)



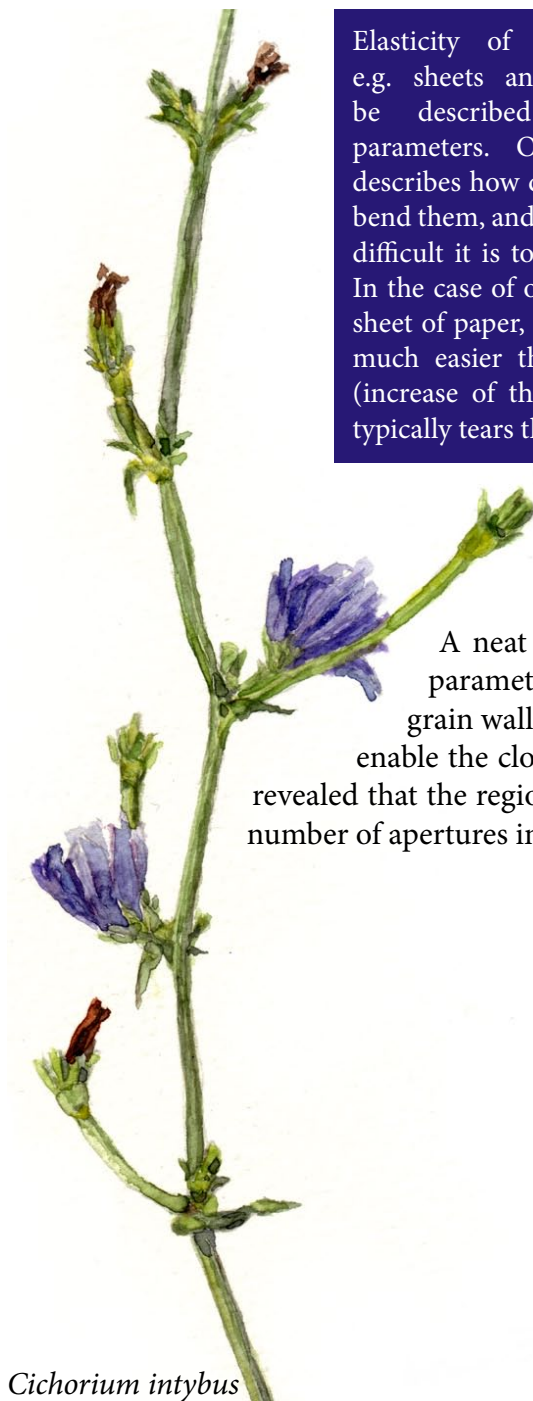
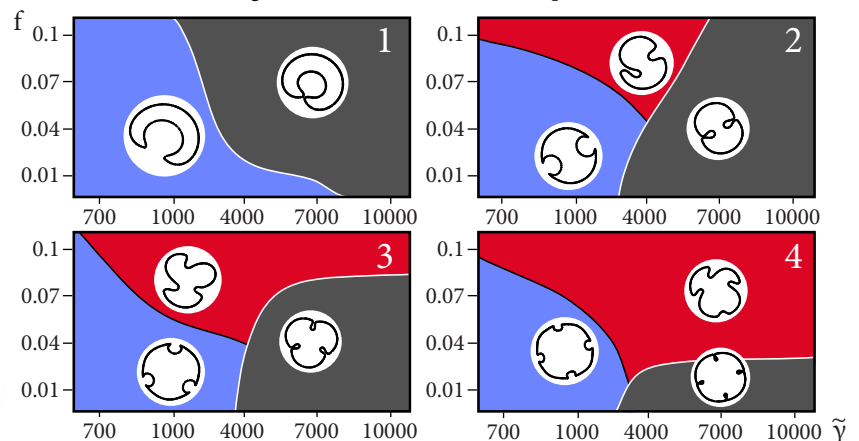
Theoretical shapes of irregularly desiccated pollen grains (7 apertures – left and 6 apertures – right). The apertures do not close and a large area of the hard part of the grain wall collapses.



Elasticity of thin objects, e.g. sheets and shells, can be described with two parameters. One of them describes how difficult it is to bend them, and the other how difficult it is to stretch them. In the case of objects like the sheet of paper, the bending is much easier than stretching (increase of the area) which typically tears the material.

A neat closure of the apertures occurs only in a limited region of elastic parameters which pertain both to the apertures and to the hard parts of the grain wall, which suggests that these parameters may be evolutionary tuned to enable the closure. Investigation of the grains with different number of apertures revealed that the region of elastic parameters which lead to regular closure shrinks as the number of apertures increases.

Grey color denotes the region of elastic parameters (relative softness of the aperture on the y-axis and combined parameter of the elasticity on the x-axis) which lead to the regular closure of the apertures. The region shrinks as the number of apertures increases (from 1 to 4).

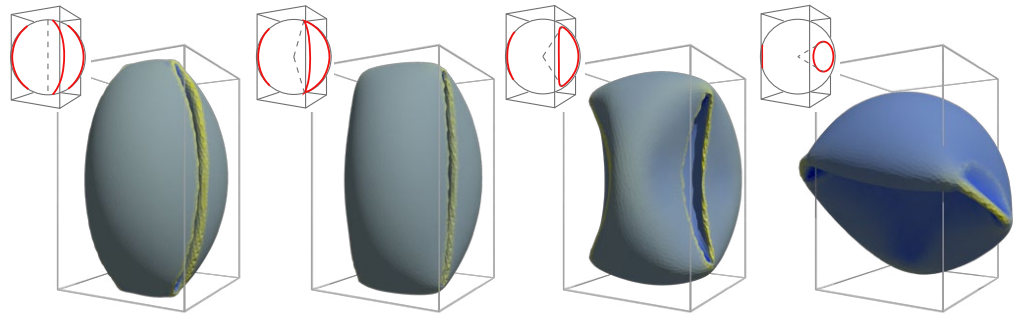


Cichorium intybus

This means that the plant species with larger number of apertures are more sensitive to the modifications of the thickness and elasticity of the grain wall and that small changes can lead to dysfunctionality of the apertures and the desiccation which destroys the pollen.

Equal volume dry grains with three apertures for different aperture shapes. The length of the aperture decreases from left to right so that the aperture of the rightmost grain is circular.

The shape and the area of apertures also influence the infolding and it was shown that long apertures which span from one pole of the grain to the other, like a garlic clove (spherical lune), lead to a more regular closure than is the case in grains with shorter, especially circular apertures.



Elongated apertures are called **colpi** and those which are nearly circular **pori**. Apertures can also be like a ring around the grain equator and in some species they spiral around the grain, connecting its poles. There are also species with apertures somewhere in between the clearly defined categories.

This study shows that although the grains with larger number of apertures germinate with larger probability, their mechanics requires a more precise tuning of the geometry and elasticity of the grain in order that it successfully closes when desiccated (the grains with smaller number of apertures close more robustly). The large number of plant species with three apertures (and not more) could thus be a result of a compromise between these two opposing trends.



The problem of a collapse of thin shells under the action of external forces or pressure is important in architecture and engineering, in particular in the design of stadiums, reservoirs, silos and underwater vehicles and structures. Homogeneous spherical shells typically collapse so that a spherical depression occurs which increases in size as the forces increase. Such shells look like deflated balls and similar shapes are indeed observed in the desiccated pollen grains without apertures or with rather small apertures.

In spite of a huge diversity of pollen grains, the study by Božič and Šiber attempts to determine some characteristics of their mechanical design which may be common for a large number of species.

The results may find application in the design of inhomogeneous elastic shells, which respond to the changes in the surrounding with a programmed deformation. Such shells could be used for encapsulation and delivery of drugs on a microscopic scale. Unlike homogeneous shells which endure the increase of the pressure all until the point of catastrophic collapse, inhomogeneous shells with designed apertures deform continuously and in a predefined fashion, depending on the magnitude of (osmotic) pressure. Soft regions in such shells would effectively guide and localize the deformation which can, in case of homogeneous spherical shells, appear in any surface point.

Microscope images in this text were adopted from the palynological database PalDat maintained by the members of the pollen research group in the Division of Structural and Functional Botany at the University of Vienna. Three-dimensional images of pollen grains are results of elastic calculations described in the work by Božič and Šiber (2020). This document was created and designed by A. Šiber in October 2020 who is also the painter of the watercolors of flowers.



Delphinium sp.